

From Le Châtelier to homeostasis.
Osmolarity, pH in biochemistry.
Buffer systems.

From the Chemistry Exam to the Final
Exam in Biochemistry
Dr. Lengyel Anna

Colligative properties

A material's property is called *colligative*, if it depends only on the number of particles, but not on the type (or chemical composition) of the particles.

Comparing the properties of a pure solvent with those of a solution:

- Vapor pressure of solution is lower
- Freezing point of solution is lower
- Boiling point of solution is higher
- Osmosis

Osmosis - the physical phenomenon

- **Osmosis** (in general): Solvent movement from the high concentration site to the low concentration site through a semipermeable membrane
- The **osmotic pressure** of a solution: the pressure that needs to be expended to prevent the ingress of water

Osmotic pressure

$$\Pi = i c R T$$

- Π = osmotic pressure
- i = van't Hoff factor
- c = molarity of **all dissolved** substances, so $i \cdot c = i_1 \cdot c_1 + i_2 \cdot c_2 + i_3 \cdot c_3 + \dots i_n \cdot c_n$
- R = universal gas constant
- T = temperature

Question

What's the difference between molarity and osmolarity (osmotic concentration)?

- Molarity (c): mol of solute in 1 dm³ solution
- Osmolarity (c_{osm}): molarity of **all dissolved** substances, so
$$i \cdot c = i_1 \cdot c_1 + i_2 \cdot c_2 + i_3 \cdot c_3 + \dots + i_n \cdot c_n$$

The difference between molarity and osmotic concentration can be illustrated by this example:

- Molarity: a 100 millimolar sodium chloride solution contains 0.1 mol NaCl per liter ($c = 0.1 \text{ mol / l} = 100 \text{ mmol / l}$).
- Osmotic concentration: in the same solution sodium chloride dissociates into the ions Na⁺ and Cl⁻, so 0.2 mol of osmotically active particles are dissolved ($c_{\text{osm}} = 0.2 \text{ osmol / l} = 200 \text{ mosmol / l}$). The actual osmotic concentration is slightly lower because not all particles dissociate and the solubility is temperature dependent.

Question

What is the normal osmotic concentration of blood plasma?

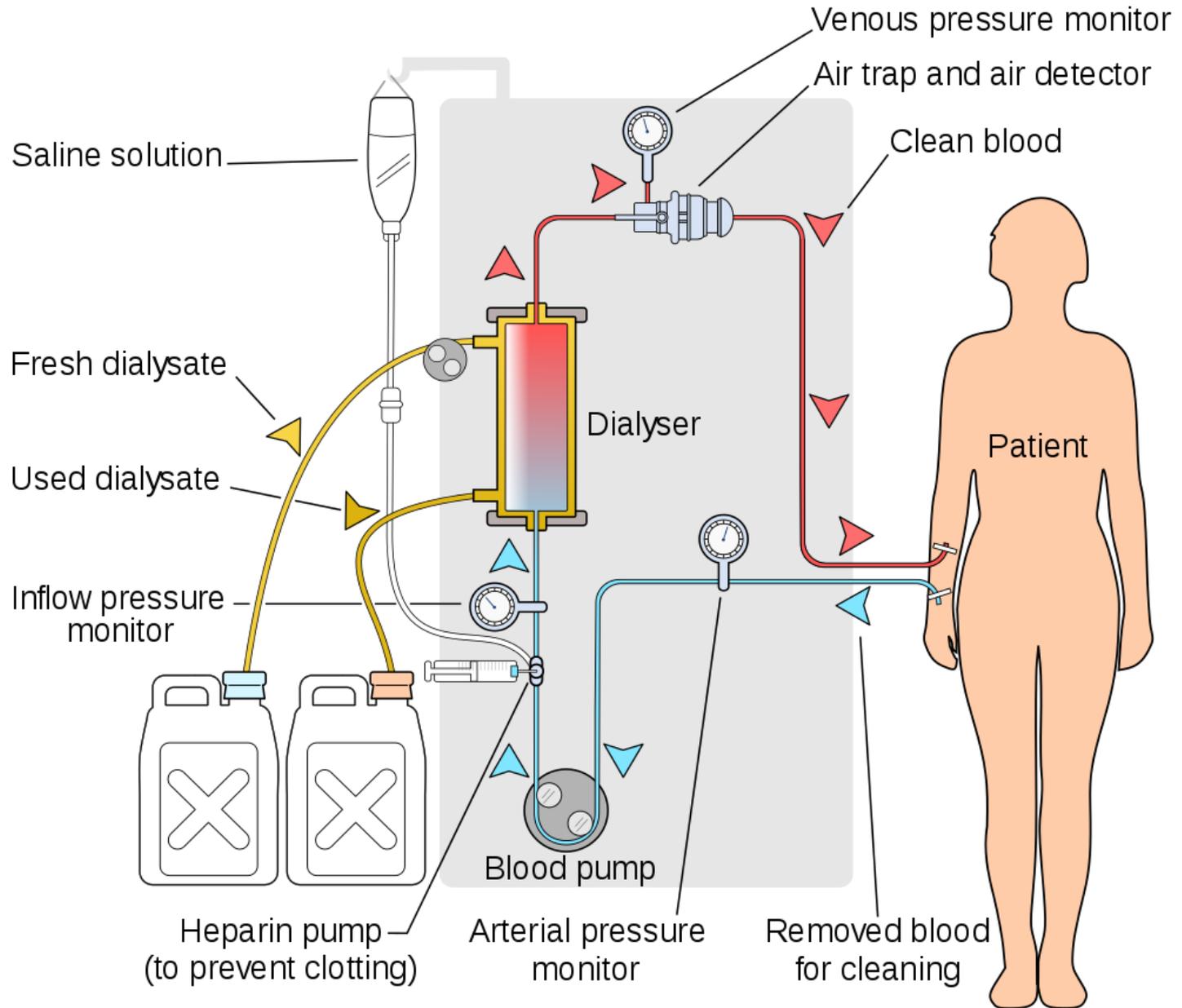
Osmolarity of blood plasma:

290 milliosmol <cBP <310 milliosmol equivalent to ~ 150 mM NaCl (0.9%)

(also known as isotonic or physiological saline)

Milliosmol = mOsm = mosmol/l

Hemodialysis



Question

Compared to water, the environment on the inside of the blood cell could best be described as:

- a. hypertonic
- b. ginandtonic
- c. isotonic
- d. hypotonic

Question

Solution	Hypotonic	Isotonic	Hypertonic
0,3M Sodium chloride			
0,05M Glucose			
0,1M Calcium chloride			
0,1M Sodium phosphate			
0,15M Urea			
0,15M Potassium chloride			
0,1M Mannose			
0,1M Magnesium chloride			
0,3M Lactate			
0,1M Ethanol			

Question

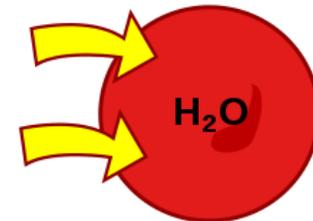
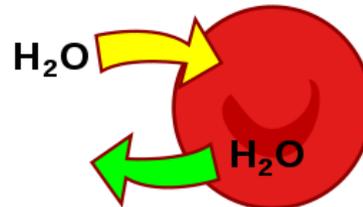
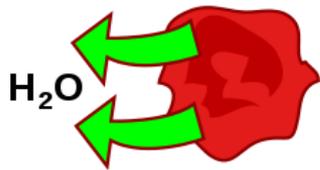
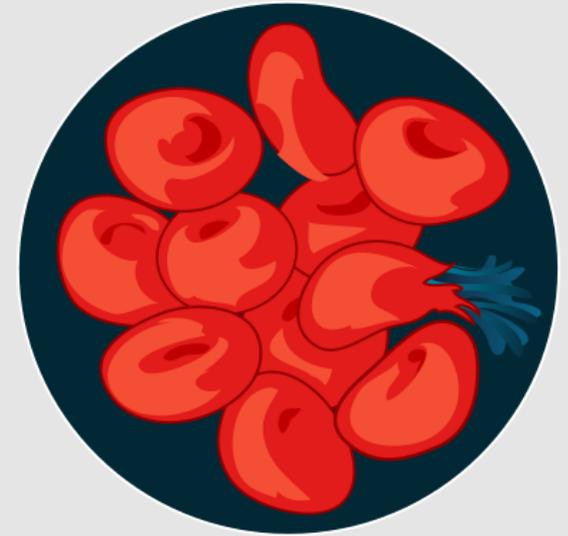
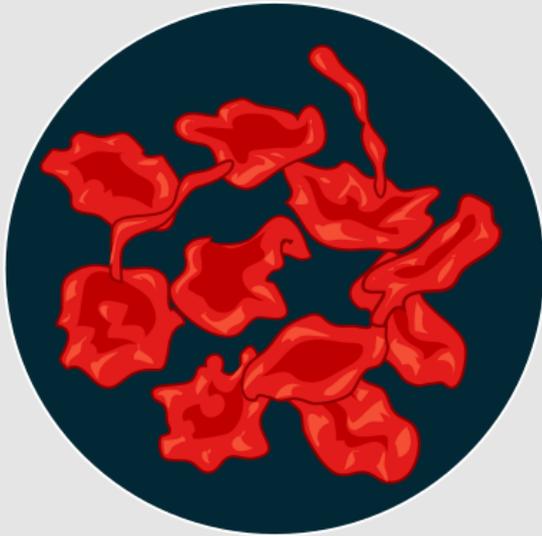
Isotonic saline solutions are preferred over hypertonic or hypotonic saline solutions when treating dehydration because _____.

- A.** Isotonic saline solutions do not cause cells to shrink from further dehydration, nor do they cause them to swell from over-hydration.
- B.** Isotonic saline solutions cause cells to shrink, making it more difficult to over-hydrate them.
- C.** Isotonic saline solutions cause cells to swell, making it more difficult to dehydrate them again.
- D.** Isotonic saline solutions are not preferred over any other saline solution.

Hypertonic

Isotonic

Hypotonic



Question

What would happen if you gave a patient an IV of pure water?

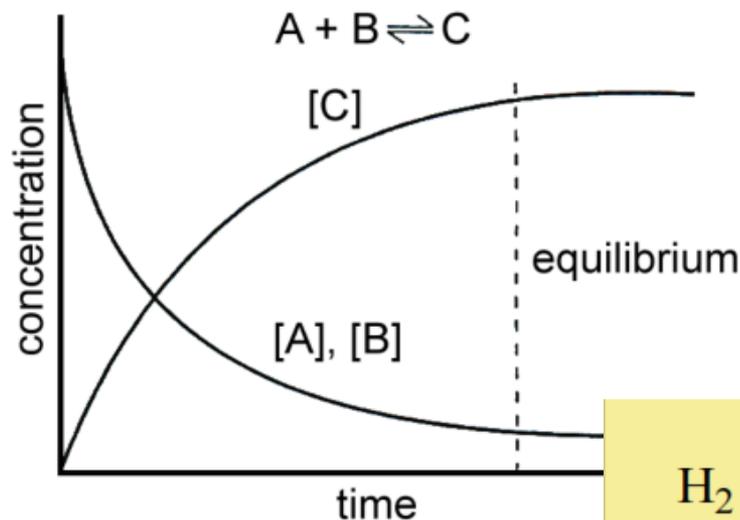
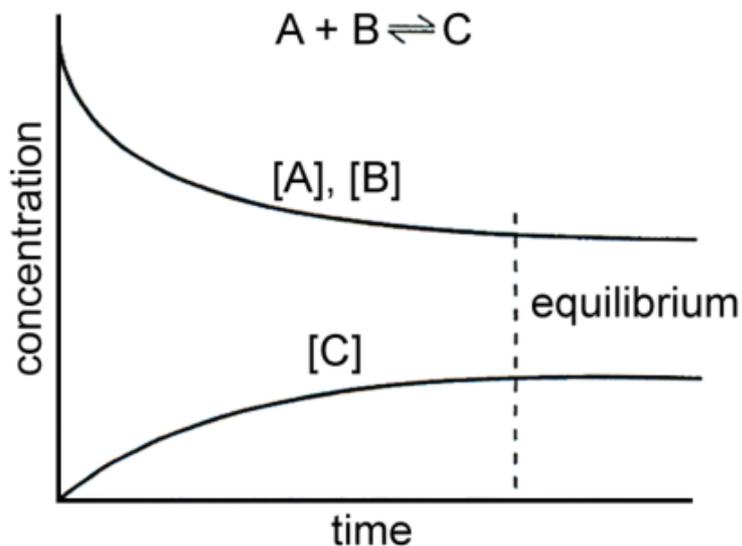
- a. Their blood cells would shrink.
- b. Their blood cells would burst.
- c. The patient would slowly become rehydrated.
- d. I would be promoted for my outstanding level of medical care.

Question

In the scenario described in the previous question, which way would water be moving?

- a. into the blood cells
- b. out of the blood cells
- c. both into and out of, but with no net change
- d. water would not be moving

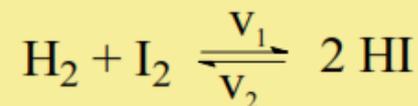
Chemical equilibria



Equilibrium is on the side of the reactants or on the side of the product.

Reversible, dynamic, the reaction rates of the forward and the reverse reaction are the same.

Mass action law gives the value of the equilibrium constant (K).



$$V_1 = k_1 \cdot [\text{H}_2] [\text{I}_2]$$

$$V_2 = k_2 \cdot [\text{HI}]^2$$

$$\text{At equilibrium : } V_1 = V_2$$

$$k_1 \cdot [\text{H}_2] [\text{I}_2] = k_2 \cdot [\text{HI}]^2$$

$$K = \frac{k_1}{k_2} = \frac{[\text{HI}]^2}{[\text{H}_2][\text{I}_2]}$$

The Equilibrium Law

Le-Châtelier's principle

If a system in equilibrium experiences a stress, a net reaction proceeds in the direction that relieves the stress.

Factors affecting the equilibrium:

- Change in concentration
- Change in pressure (only gas reactions)
- Change in temperature

Question



- What's the type of this equilibrium?
- What happens with the equilibrium if
 - a. we increase the pressure?
 - b. we remove NH_3 ?
 - c. we remove N_2 ?
 - d. we add catalyst?



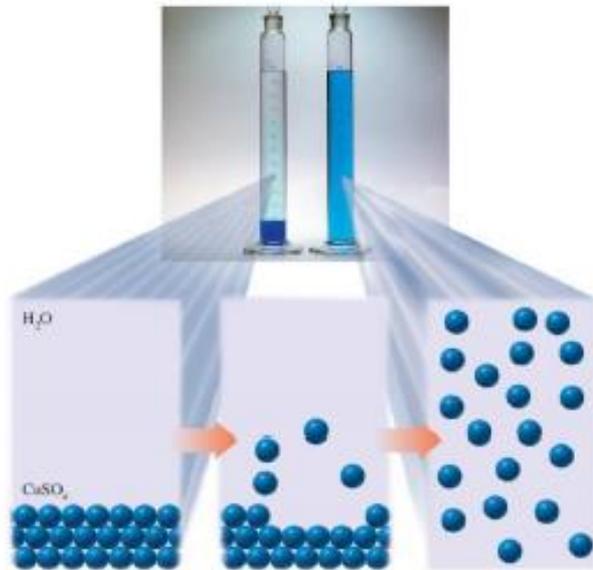
$$\Delta H^\circ = -180 \text{ kJ}$$

$[\text{O}_2] \uparrow$ When is this right?

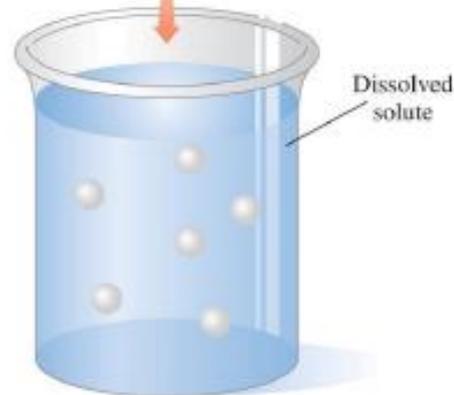
Solubility

The maximum amount of a solute that can be dissolved in a certain amount of solvent at a given temperature. The concentration of the saturated solution at a given temperature.

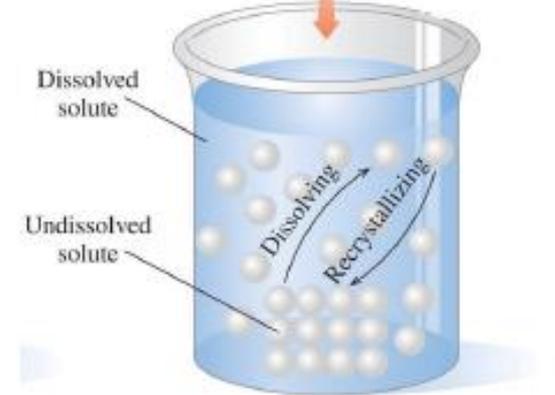
Depends on temperature and on pressure (by gases).



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Not saturated solution



Saturated solution

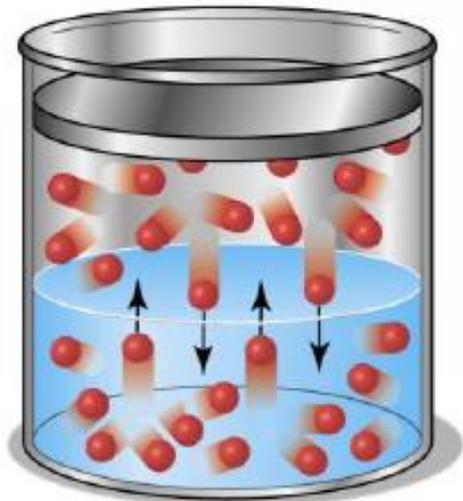
Effect of pressure on the solubility of gases

Henry's Law

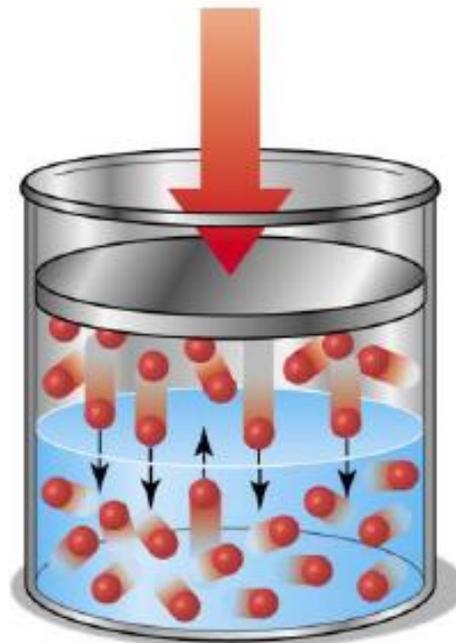
$$\text{Solubility} = k \cdot p$$

p: partial pressure of the gas over the solution

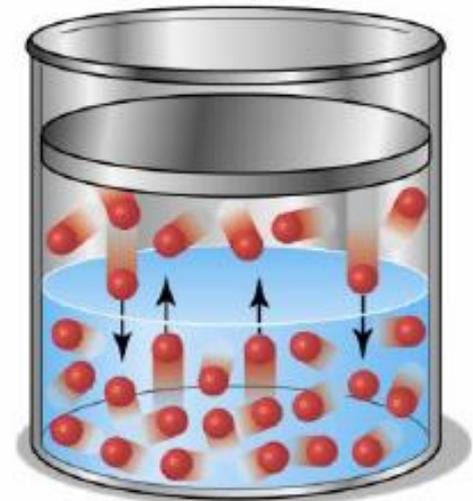
k: constant,
depending on quality and on temperature



Equilibrium



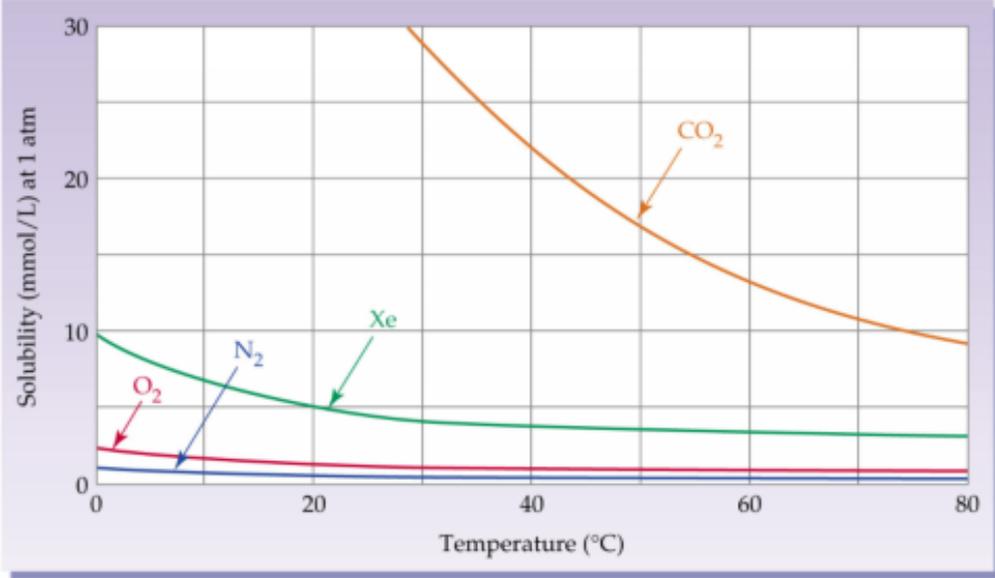
Pressure increased



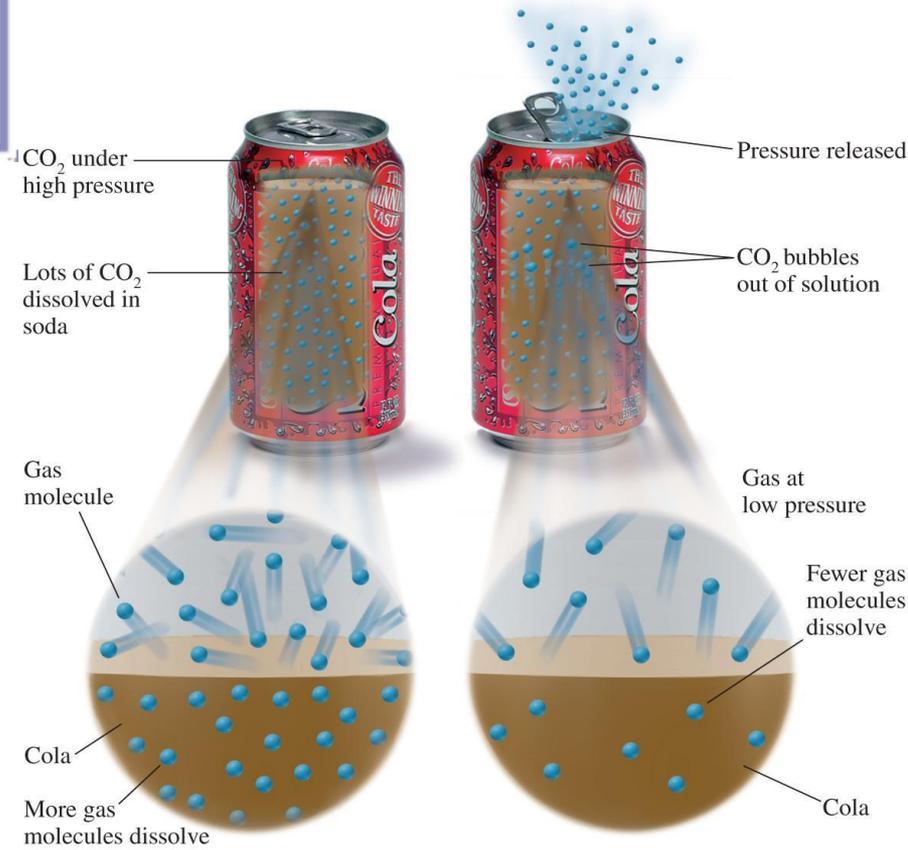
New equilibrium

Solubility of gases: inversely proportional with the temperature

Question

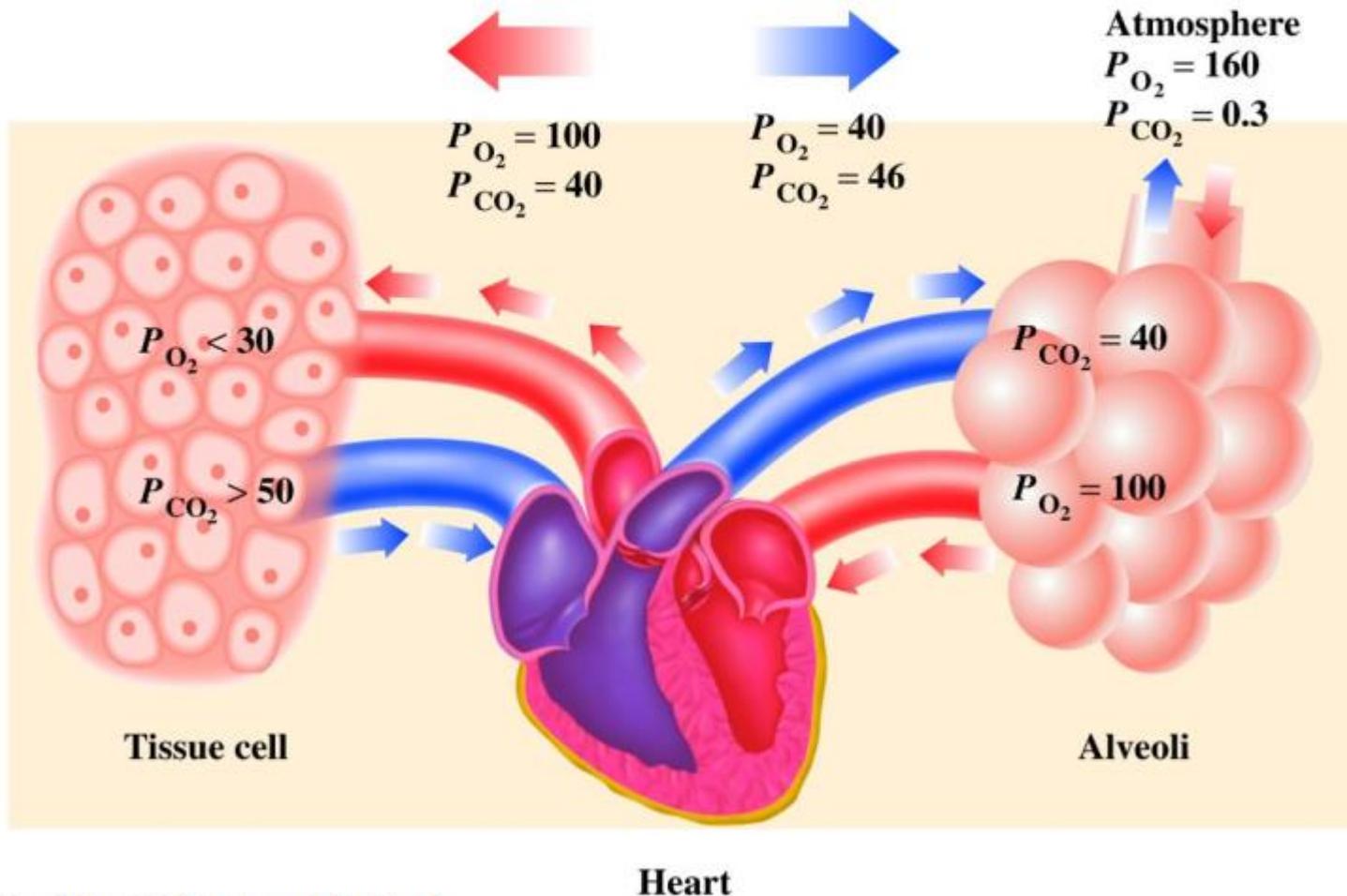


Why do we drink coke, beer, champagne, etc. cold, but tea hot?



- In the lungs, O_2 enters the blood, while CO_2 from the blood is released.
- In the tissues, O_2 enters the cells, which release CO_2 into the blood.

Blood Gases



Electrolytes

Electrolytes: ionic or polar covalent compounds.

Electrolytic dissociation: release of ions of an electrolyte when dissolved in water.
(From the ion lattice, ion-dipole attractions, hydration shell)

Strong electrolytes: dissociate in large extent (70-100%), that's why they have electrical conductivity. e.g. strong acids, strong bases, water-soluble salts
In solid state there are no ions that could move freely.

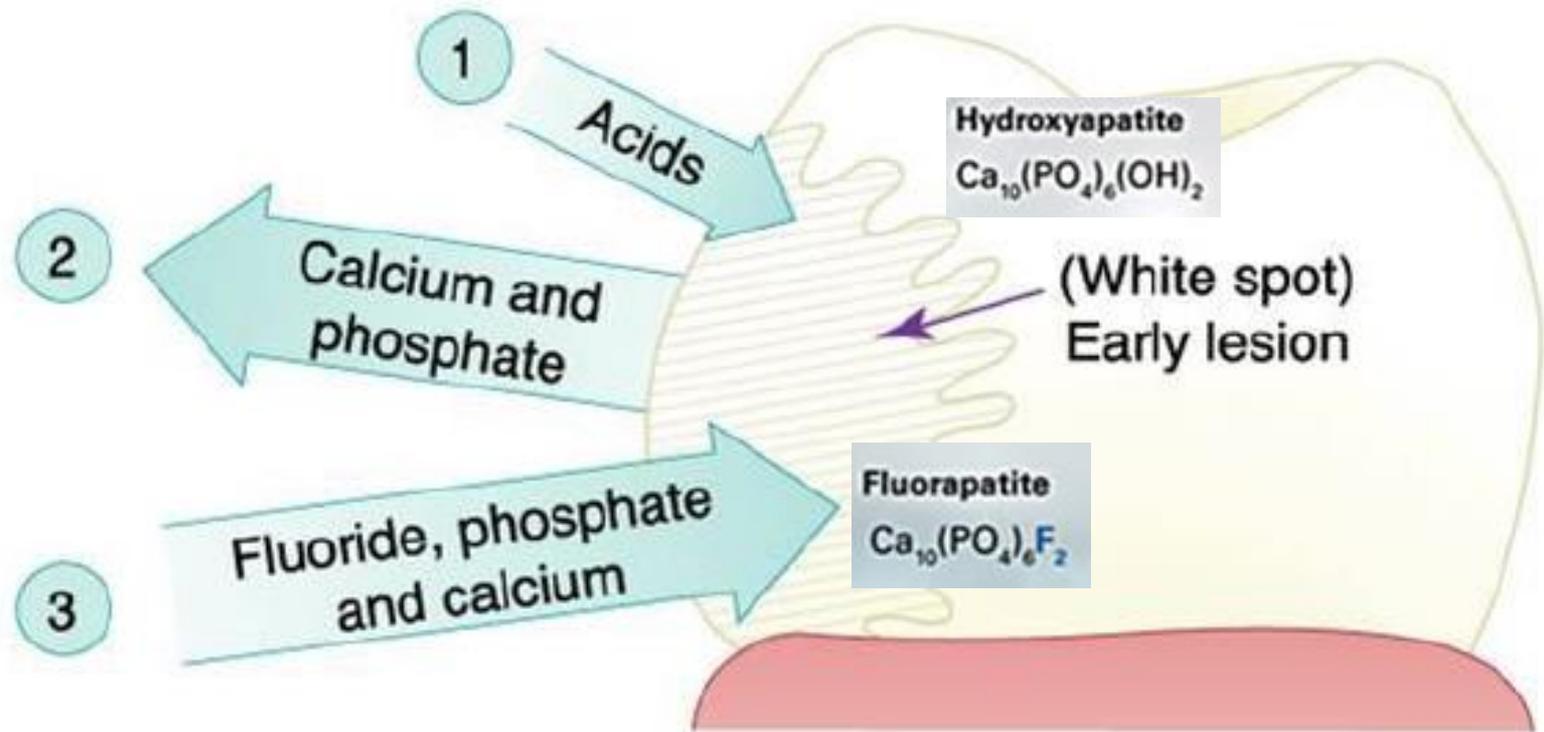
Weak electrolytes: dissociate in slight extent, only partially, so they predominantly exist as molecules in solutions. e.g. weak acids, weak bases

Non-electrolytes: in solutions do not form ions, therefore they do not conduct electricity at all. e.g. ethanol, sucrose, urea

Solubility: the concentration of the saturated solution means the solubility of the given substance.

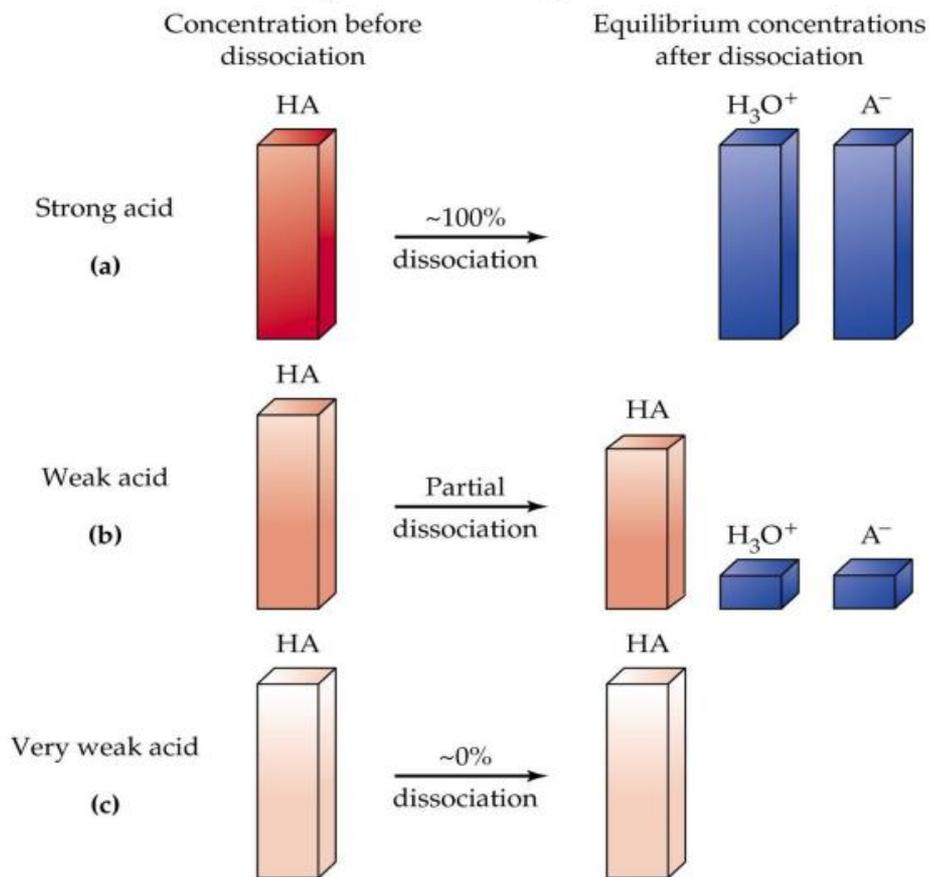
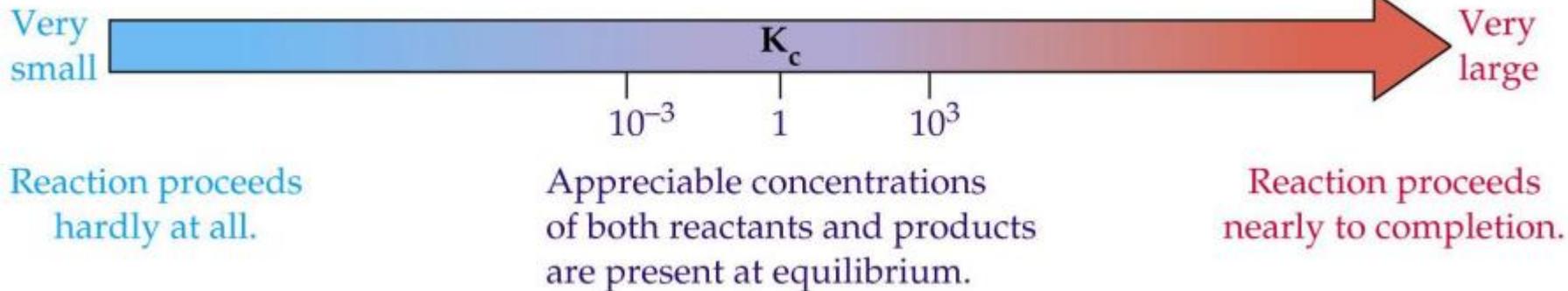
It depends on the **temperature**.

Ionic compounds with basic anions have better solubility at a **lower pH**.



1. The tooth is attacked by acids in plaque and saliva.
2. Calcium and phosphate dissolve from the enamel in the process of demineralization.
3. Fluoride, phosphate and calcium re-enter the enamel in a process called remineralization.

Strong or weak?



Weak electrolytes

While they do not dissociate completely in the solution, the dissolved molecule is in equilibrium with the ions.

Degree of dissociation (α)

A fraction of the total amount of the electrolyte that is dissociated into ions. (Sometimes given in percentage.) It always depends on the concentration.

α = concentration of dissociated moles/concentration of total moles

If **HA** is a weak acid, and the initial concentration of the acid is c (mol/dm³), the dissociation constant (K_a) is equal to:

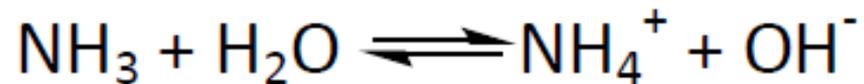
$$\frac{\alpha^2 \cdot c}{1 - \alpha}$$

where $1 - \alpha$ is approx. 1

Law of mass action

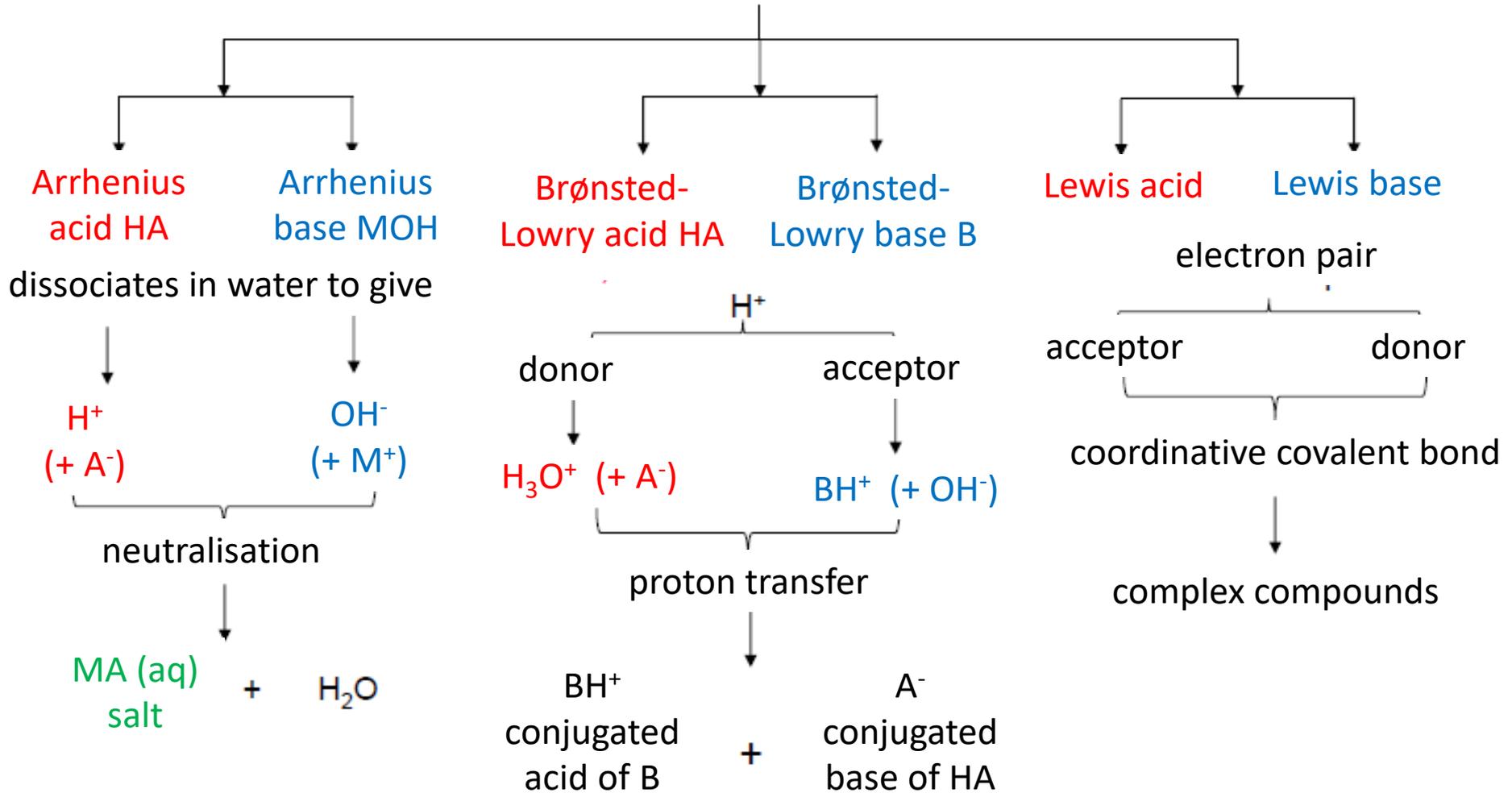


$$K_a = \frac{[\text{HCOO}^-][\text{H}^+]}{[\text{HCOOH}]}$$



$$K_b = \frac{[\text{NH}_4^+][\text{OH}^-]}{[\text{NH}_3]}$$

ACIDS AND BASES



They can be:

- Strong or weak
- Monoprotic or polyprotic

Arrhenius theory:

Acids dissociate in water to produce H^+ or H_3O^+ ions



Bases dissociate in water to generate OH^- ions



Brønsted-Lowry theory:

Acids are proton donor molecules or ions e.g. NH_4^+ , HSO_4^-

Bases are proton acceptor substances e.g. CN^-

Acid-base reactions are proton transfer reactions.

An acid losing a proton produces a *conjugated base*, a base accepting a proton yields a *conjugated acid*.

Lewis theory: (dative or coordinative bond)

Lewis acids: electron-pair acceptors

Lewis bases: electron-pair donors

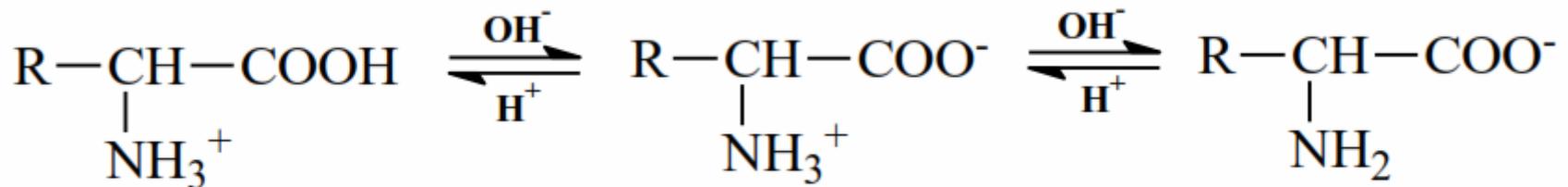
Amphoterism

Definition: they can behave both as an acid and as a base depending on the partner.

Inorganic amphoteric compounds

Amphoteric oxides: oxides not soluble in water, but soluble in acids and bases (e.g. Al_2O_3 , ZnO).

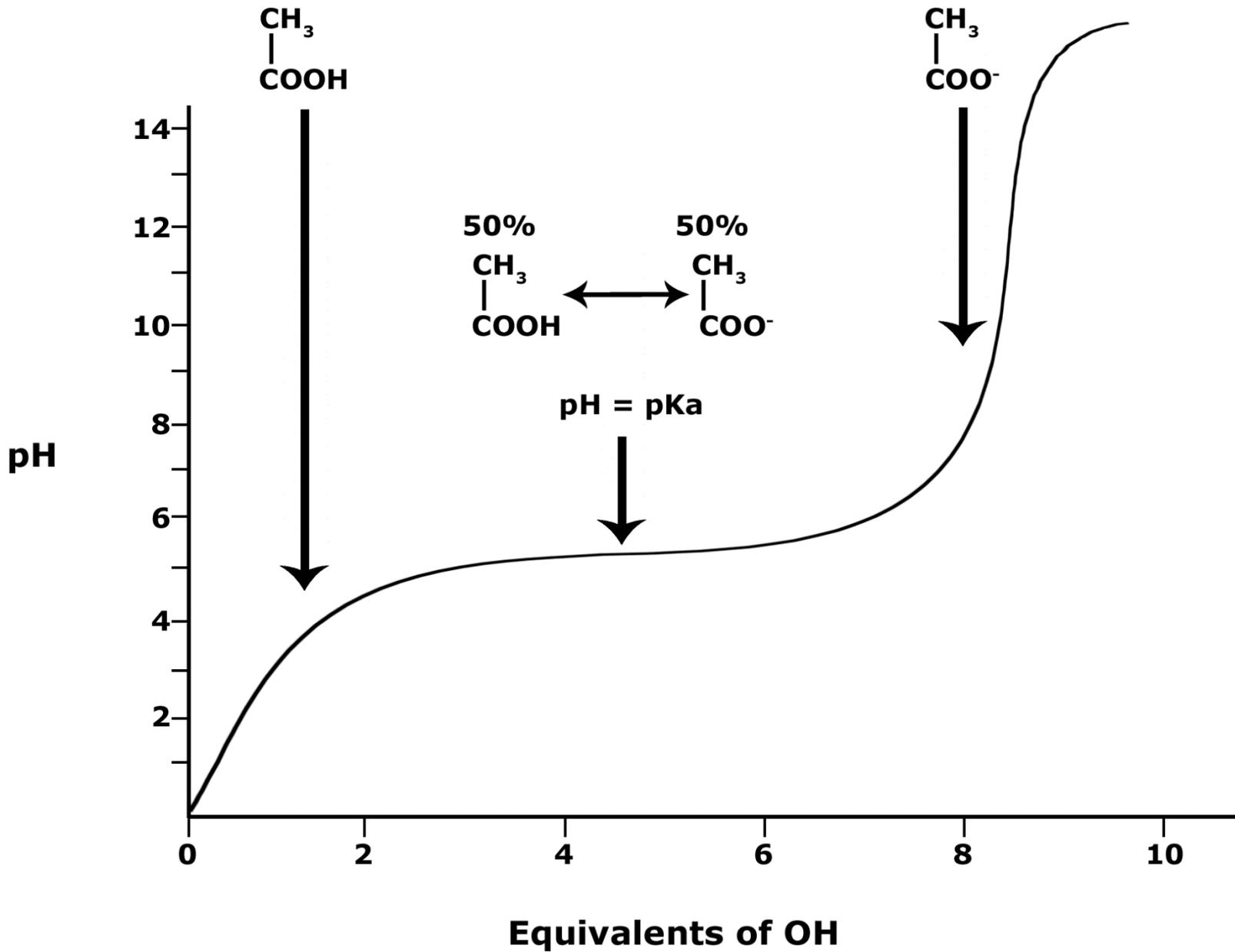
Amphoteric character of amino acids

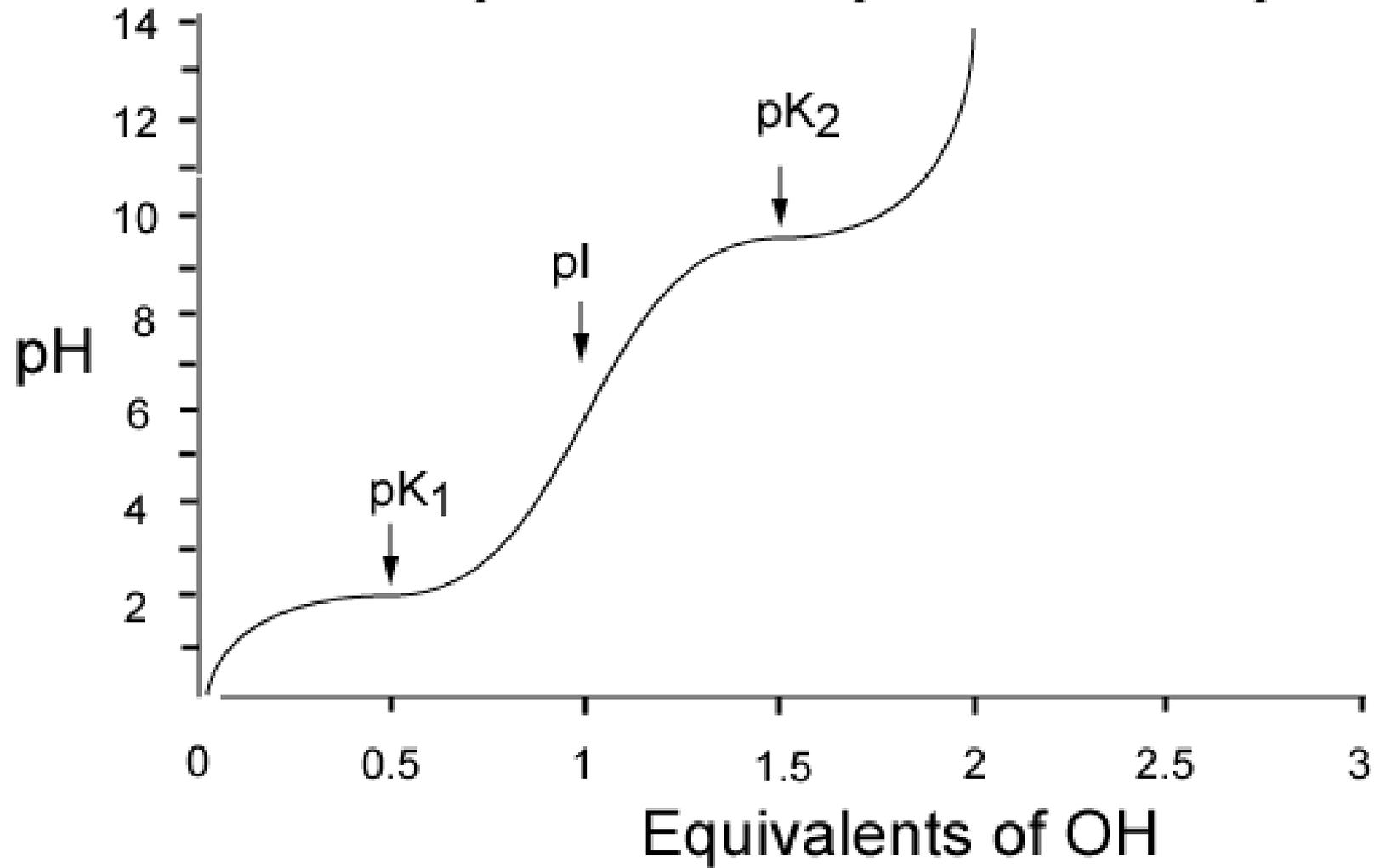
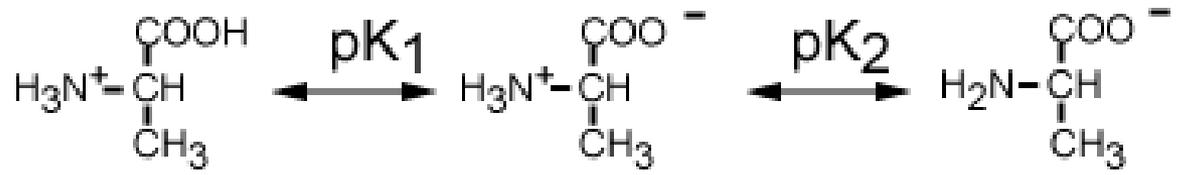


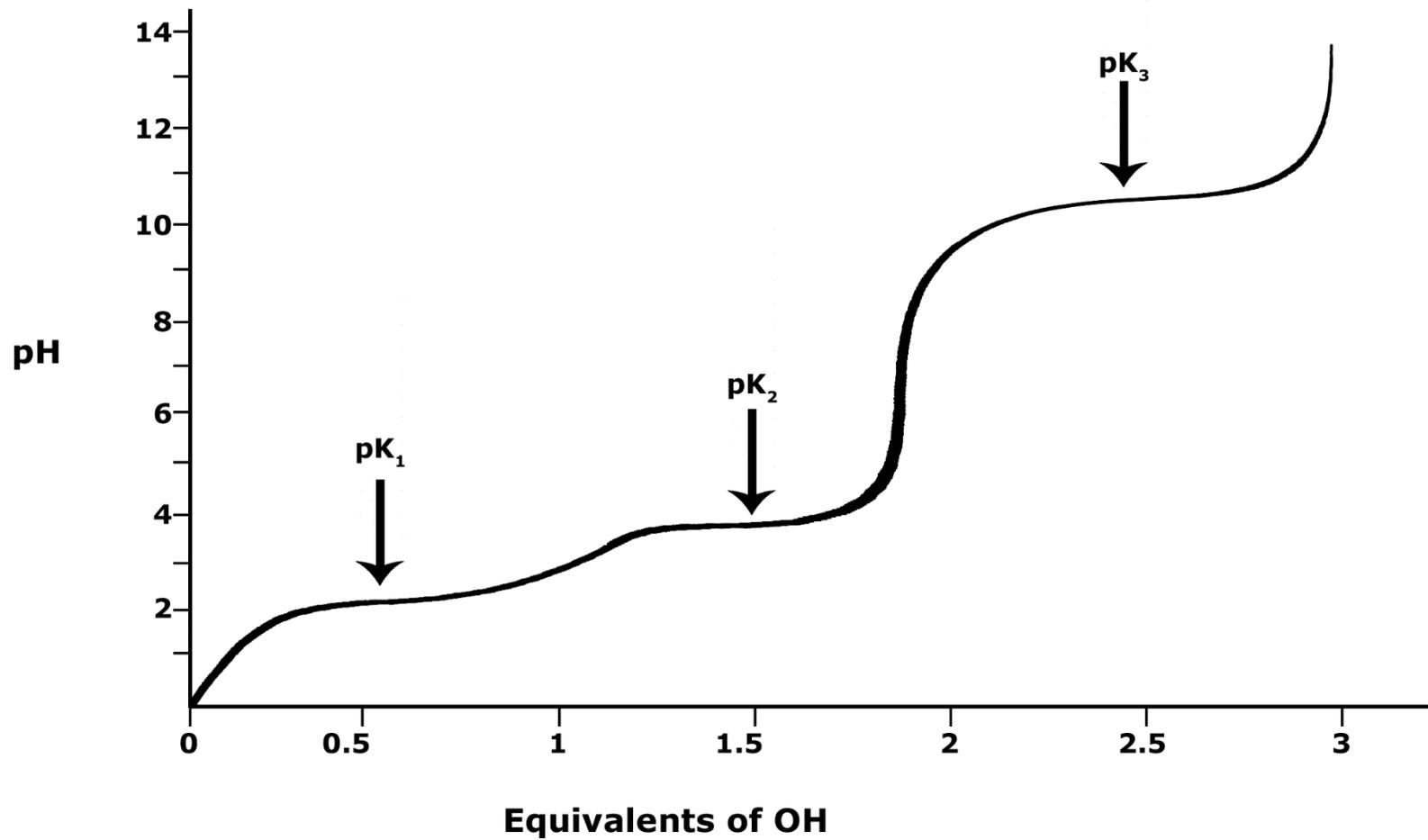
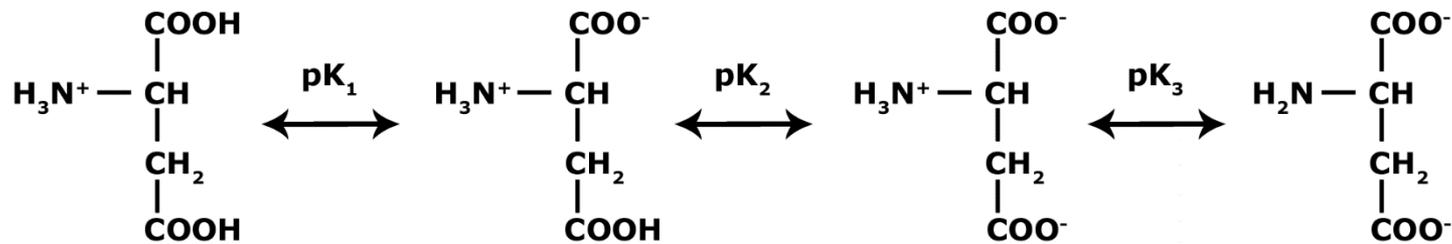
cation
(acidic medium)

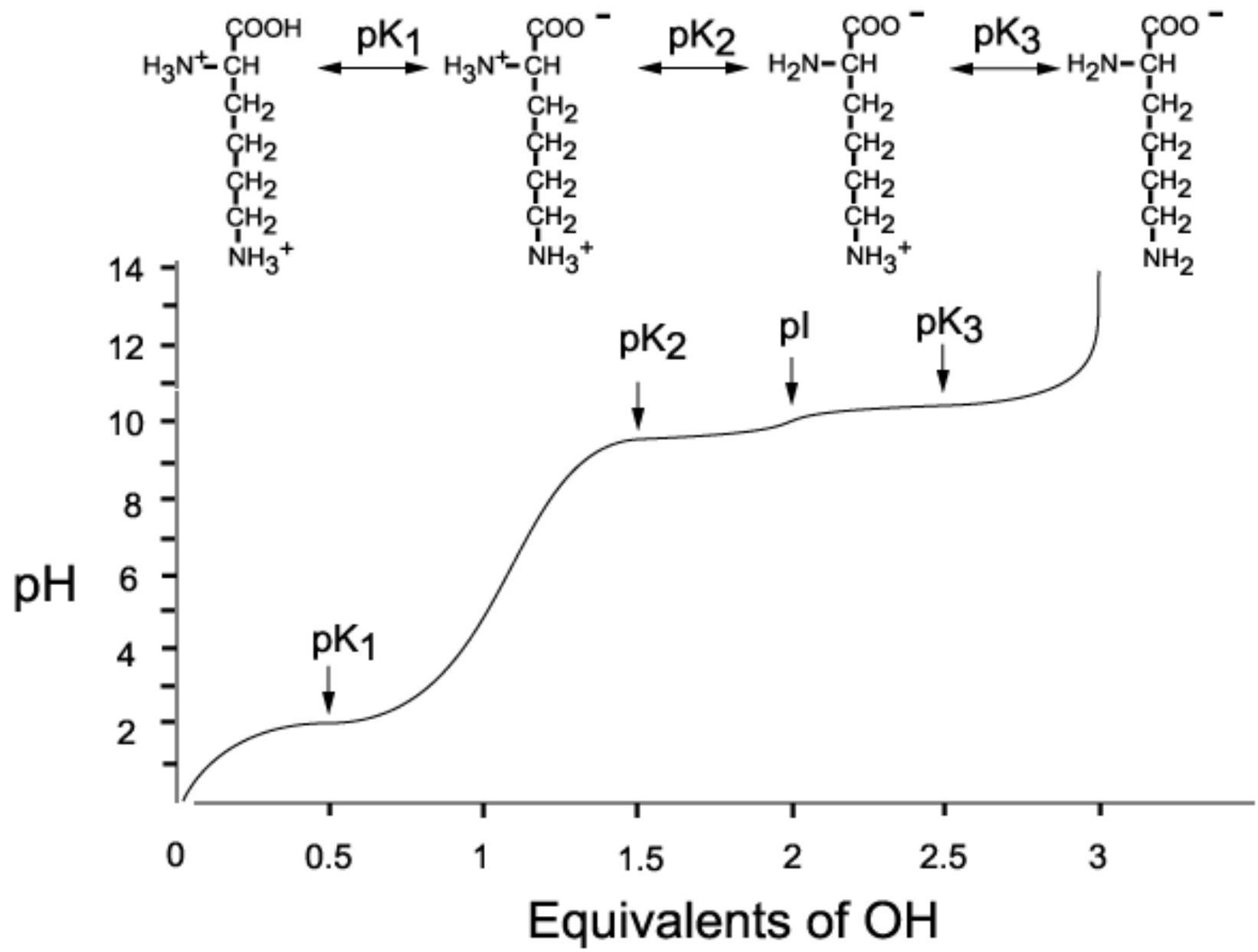
zwitterion
(~ neutral medium)

anion
(basic medium)



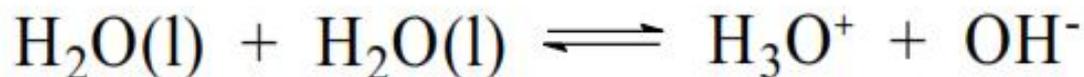
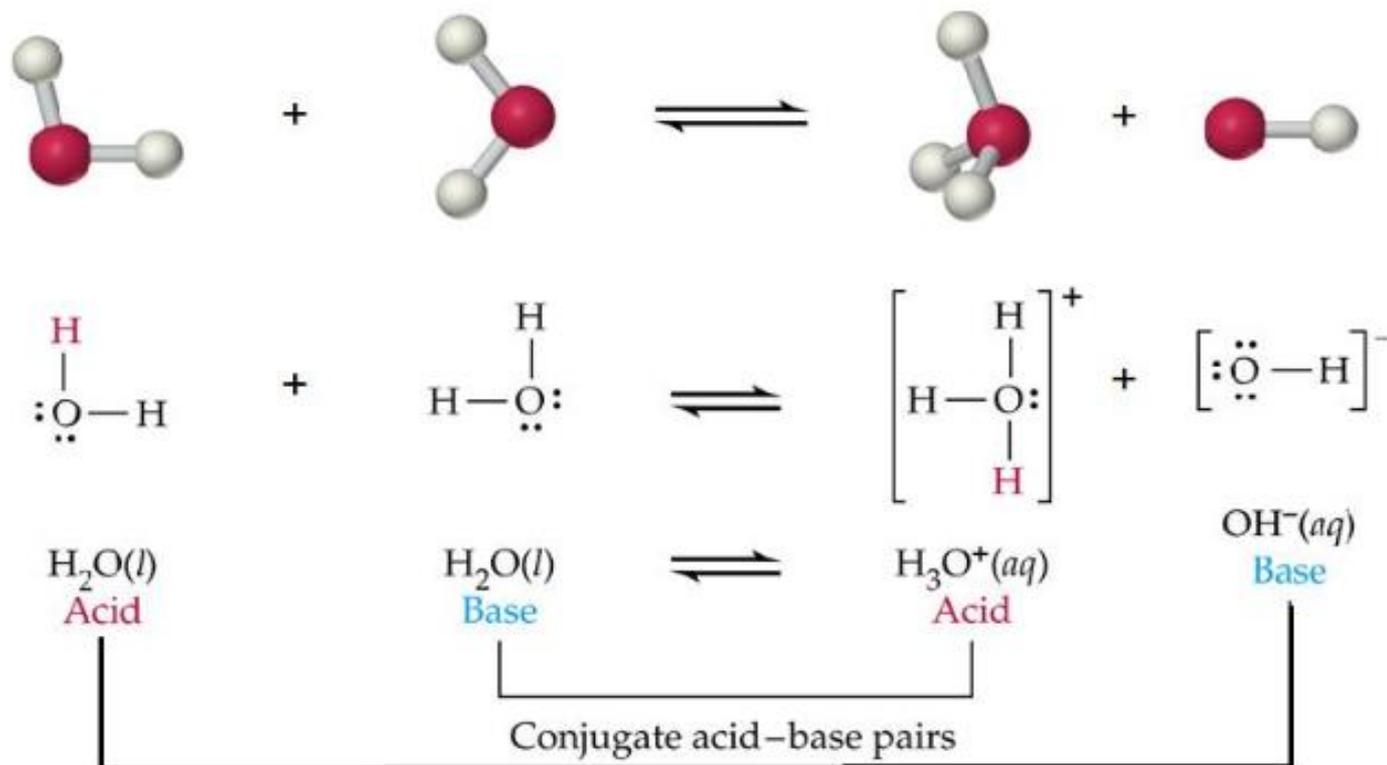




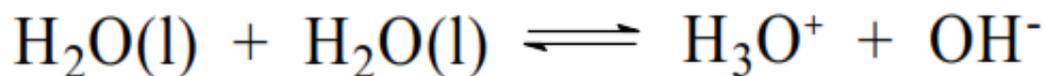


The self-ionization of water

The water behaves as a very weak electrolyte showing small conduction. It is an **amphoteric compound** having **both acidic and basic properties**.



Dissociation of water



Applying the **law of mass action**:

$$K = \frac{[\text{H}_3\text{O}^+][\text{OH}^-]}{[\text{H}_2\text{O}]^2} = 1,8 \times 10^{-16}$$

Because the water is a very weak electrolyte, the concentration of the water remains constant, it is incorporated into the equilibrium constant.

Ion-product constant of water

$$K_w = [\text{H}^+][\text{OH}^-] = 1.00 \cdot 10^{-14} \quad (\text{at } 25 \text{ } ^\circ\text{C})$$

The temperature influences the dissociation.

In pure water: $[\text{H}^+] = [\text{OH}^-] = 1.00 \cdot 10^{-7} \text{ mol/dm}^3$

The pH and the pOH

pH is the negative logarithm of the hydronium ion concentration.

$$\text{pH} = -\log [\text{H}_3\text{O}^+]$$

$$\text{pH} = -\log [\text{H}^+] = \log \frac{1}{[\text{H}^+]}$$

pOH is the negative logarithm of the hydroxide ion concentration

$$\text{pOH} = -\log [\text{OH}^-] = \log \frac{1}{[\text{OH}^-]}$$

The connection between the pH and pOH is the following:

$$K_w = [\text{H}^+] [\text{OH}^-] = 1.00 \cdot 10^{-14} \quad (\text{at } 25 \text{ } ^\circ\text{C})$$

$$\text{p}K_w = -\log K_w = 14.00$$

$$\text{p}K_w = \mathbf{\text{pH} + \text{pOH} = 14.00}$$

In neutral solution:

$$\mathbf{\text{pH} = \text{pOH} = 7.00}$$

Hydrolysis of salts

Definition: the reaction of ions with water to yield protons or hydroxide ions.

- Salts of strong acids and strong bases have always a neutral solution. (pH=7). E.g. NaCl, KCl
- Salts derived from a strong base and a weak acid (e.g. $\text{CH}_3\text{-COONa}$) mean anion hydrolysis in the solution (pH>7). The weak acid is protonated, so the OH^- concentration increases and the solution is basic.
- Salts derived from a weak base and a strong acid (e.g. NH_4Cl) mean cation hydrolysis in the solution (pH<7). The weak base is deprotonated, the H^+ concentration increases, the solution is acidic.
- Salts derived from a weak base and a weak acid (e.g. NH_4NO_2) mean both anion and cation hydrolysis simultaneously in the solution. The pH value is determined by the relative magnitude of the K values. (In this case K_a of NH_4^+ ions is larger than K_b of NO_2^- ions, so the solution is acidic.)

General expression for acidic buffers:

$$[H^+] = K_a \cdot \frac{[HA]}{[A^-]}$$

$$-\log [H^+] = -\log K_a + \log \frac{[A^-]}{[HA]}$$

$$pH = pK_a + \log \frac{[A^-]}{[HA]}$$

The equation above is the **Henderson-Hasselbalch equation** describing the **pH value of an acidic buffer system, which depends only on the ratio of the components.**

$$K_b = \frac{[NH_4^+][OH^-]}{[NH_3]}$$

$$[OH^-] = K_b \frac{[NH_3]}{[NH_4^+]} \quad pOH = pK_b + \log \frac{[NH_4^+]}{[NH_3]}$$

$$pOH = pK_b + \log \frac{[HB^+]}{[B]} \quad \begin{array}{l} \mathbf{HB^+} = \text{conjugated} \\ \mathbf{acid} \\ \mathbf{B} = \text{base} \end{array}$$

Henderson-Hasselbalch equation for basic buffers.

The most important buffer systems in humans:

- Phosphate buffer in the intracellular space
- Carbonic acid / bicarbonate system in the blood and in interstitial space
- Albumin in blood plasma
- Hemoglobin in red blood cells

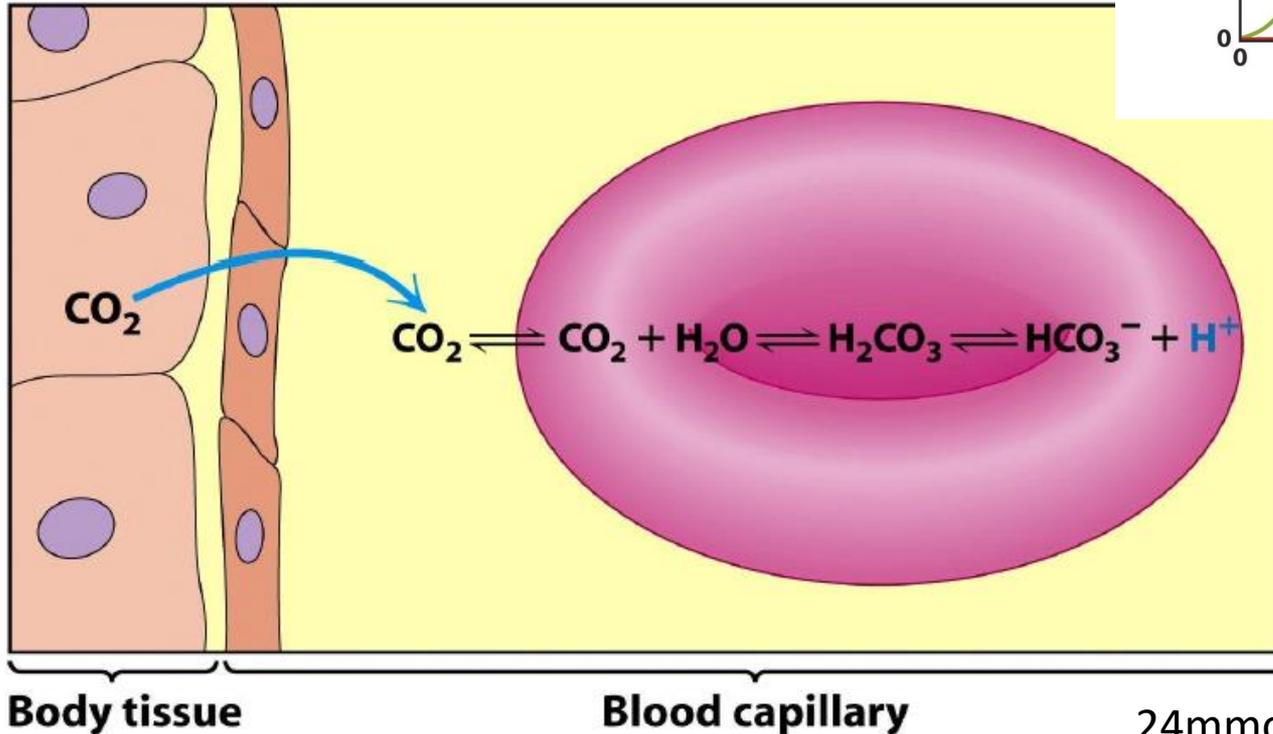
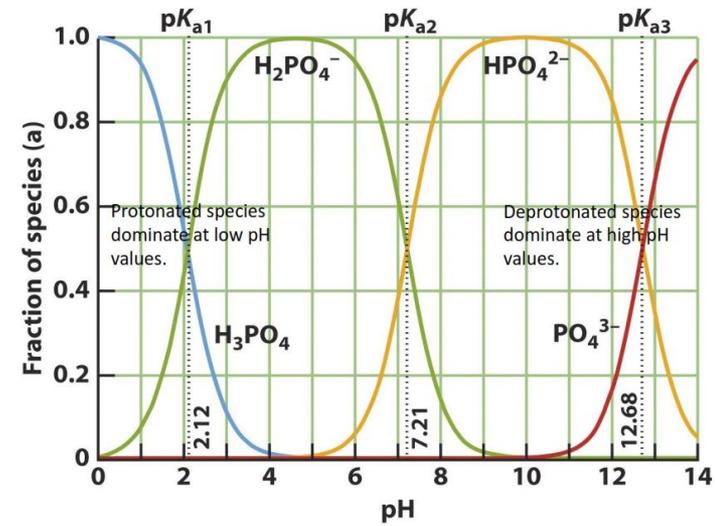


Figure 7-20
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$$pH = pK_a + \log \frac{[HCO_3^-]}{[H_2CO_3]} = 6,1 + \log \frac{[HCO_3^-]}{0,03pCO_2} = 6,1 + \log 20 = 7,4$$

Solubility of CO_2

Buffer capacity

Definition:

The amount of strong acid or strong base in moles required to shift the pH of 1l buffer solution by one unit.

Question

Which of the following buffer solutions has a higher buffer capacity against the 0.1 M NaOH solution?

- a. 0,1 M Na_2HPO_4 and 0,15 M NaH_2PO_4
- b. 0,15 M Na_2HPO_4 and 0,1 M NaH_2PO_4

Question

Most of the fluids produced by the human body are ...

- a. very acidic
- b. in the neutral range
- c. very basic
- d. very alkaline

Question

The difference between a strong acid and a weak acid involves...

- a. how much of it dissociates in water
- b. how much it synthesizes in water
- c. how much of it changes into water
- d. hours spent lifting weights

Question

Each number on the pH scale represents a _____ change in H⁺ concentration.

- a. one point
- b. tenfold
- c. hundredfold
- d. ten thousandfold
- e. negative

Question

A salt is:

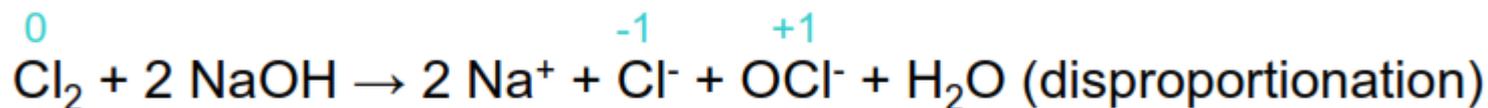
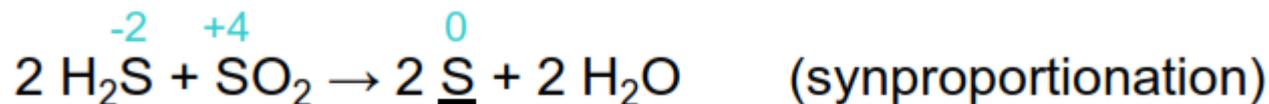
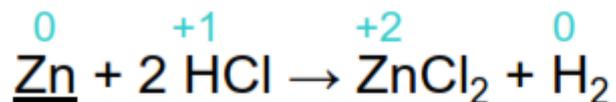
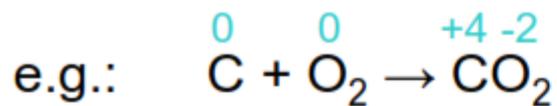
- a. A compound that dissociates in water and releases hydrogen ions (H^+).
- b. An atom that dissociates in water and forms cations and anions.
- c. A compound that dissociates in water and releases hydroxyl ions (OH^-).
- d. A compound that dissociates in water and produces cations or anions other than H^+ or OH^- .

Question

The body's bicarbonate buffer system functions to keep the pH of the blood neutral. Which two organs/systems are primarily involved in keeping this buffer system balanced?

- a. lungs, liver
- b. digestive, circulatory
- c. digestive, respiratory
- d. kidneys, lungs
- e. immune, circulatory

Redox reactions: chemical reactions with electron transfer



Oxidation: loss of electrons (increase of oxidation number)

Reduction: take-up of electrons (decrease of oxidation number)

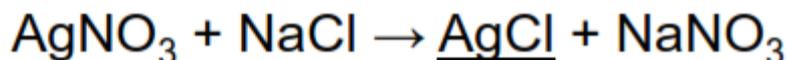
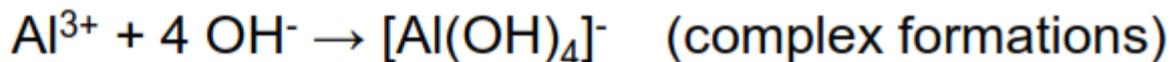
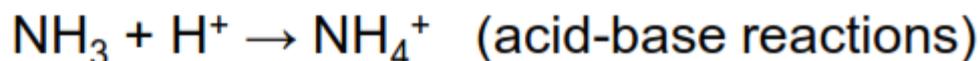


TABLE 12.1 Standard Potentials at 25°C*

More p**o**sitive \longrightarrow **O**xidizes more negative

Species	Reduction half-reaction	E° (V)
strong oxidizing agents		
Ce^{4+}/Ce^{3+}	$F_2(g) + 2 e^- \longrightarrow 2 F^-(aq)$	+1.61
$MnO_4^-, H^+/Mn^{2+}, H_2O$	$Au^+(aq) + e^- \longrightarrow Au(s)$	+1.51
Cl_2/Cl^-	$Ce^{4+}(aq) + e^- \longrightarrow Ce^{3+}(aq)$	+1.36
$Cr_2O_7^{2-}, H^+/Cr^{3+}, H_2O$	$MnO_4^-(aq) + 8 H^+(aq) + 5 e^- \longrightarrow Mn^{2+}(aq) + 4 H_2O(l)$	+1.33
$O_2, H^+/H_2O$	$Cl_2(g) + 2 e^- \longrightarrow 2 Cl^-(aq)$	+1.23;
	$Cr_2O_7^{2-} + 14 H^+(aq) + 6 e^- \longrightarrow 2 Cr^{3+}(aq) + 7 H_2O(l)$	+0.82 at pH
	$O_2(g) + 4 H^+(aq) + 4 e^- \longrightarrow 2 H_2O(l)$	+1.09
		+0.96
Br_2/Br^-	$Br_2(l) + 2 e^- \longrightarrow 2 Br^-(aq)$	+0.80
$NO_3^-, H^+/NO, H_2O$	$NO_3^-(aq) + 4 H^+(aq) + 3 e^- \longrightarrow NO(g) + 2 H_2O(l)$	+0.77
Ag^+/Ag	$Ag^+(aq) + e^- \longrightarrow Ag(s)$	+0.54
Fe^{3+}/Fe^{2+}	$Fe^{3+}(aq) + e^- \longrightarrow Fe^{2+}(aq)$	+0.40;
I_2/I^-	$I_2(s) + 2 e^- \longrightarrow 2 I^-(aq)$	+0.82 at pH
$O_2, H_2O/OH^-$	$O_2(g) + 2 H_2O(l) + 4 e^- \longrightarrow 4 OH^-(aq)$	+0.34
		+0.22
		0, by definit
Cu^{2+}/Cu	$Cu^{2+}(aq) + 2 e^- \longrightarrow Cu(s)$	-0.04
$AgCl/Ag, Cl^-$	$AgCl(s) + e^- \longrightarrow Ag(s) + Cl^-(aq)$	-0.08
H^+/H_2	$2 H^+(aq) + 2 e^- \longrightarrow H_2(g)$	-0.13
Fe^{3+}/Fe	$Fe^{3+}(aq) + 3 e^- \longrightarrow Fe(s)$	-0.14
$O_2, H_2O/HO_2^-, OH^-$	$O_2(g) + H_2O(l) + 2 e^- \longrightarrow HO_2^-(aq) + OH^-(aq)$	-0.44
Pb^{2+}/Pb	$Pb^{2+}(aq) + 2 e^- \longrightarrow Pb(s)$	-0.76
Sn^{2+}/Sn	$Sn^{2+}(aq) + 2 e^- \longrightarrow Sn(s)$	-0.83;
Fe^{2+}/Fe	$Fe^{2+}(aq) + 2 e^- \longrightarrow Fe(s)$	-0.42 at pH
Zn^{2+}/Zn	$Zn^{2+}(aq) + 2 e^- \longrightarrow Zn(s)$	-1.66
$H_2O/H_2, OH^-$	$2 H_2O(l) + 2 e^- \longrightarrow H_2(g) + 2 OH^-(aq)$	-0.83;
		-0.42 at pH
Al^{3+}/Al	$Al^{3+}(aq) + 3 e^- \longrightarrow Al(s)$	-1.66
weak oxidizing agents	$Mg^{2+}(aq) + 2 e^- \longrightarrow Mg(s)$	-1.66
	$Na^+(aq) + e^- \longrightarrow Na(s)$	-1.66
	$K^+(aq) + e^- \longrightarrow K(s)$	-1.66
	$Li^+(aq) + e^- \longrightarrow Li(s)$	-1.66

spontaneous

non-spontaneous

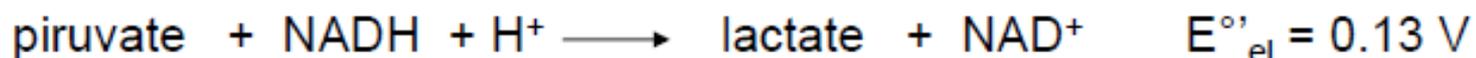
Reduced form is strongly reducing

*For a more extensive table, see Appendix 2B.

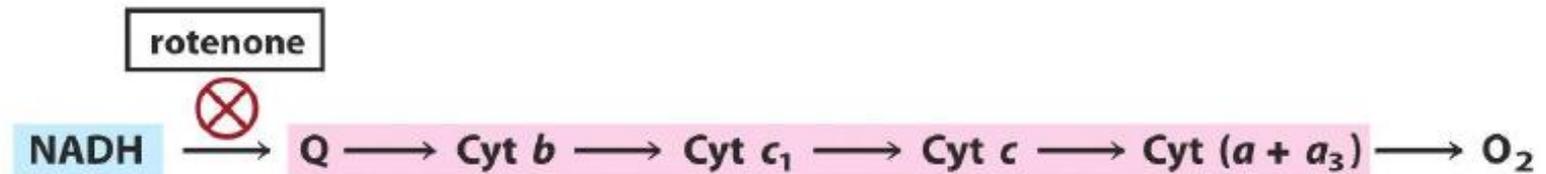
More n**e**gative \longrightarrow r**e**duces more positive

Redox processes in metabolism

redox process (ox + n e ⁻ → red)	n	ε ^{o'} (V)
½ O ₂ (g) + 2 H ⁺ + 2 e ⁻ → H ₂ O (l)	2	0.81
cytochrome-c ₁ (Fe ³⁺) → cytochrome-c ₁ (Fe ²⁺)	1	0.22
dehydroascorbate → ascorbate	2	0.08
fumarate → succinate	2	0.03
piruvate → lactate	2	- 0.19
acetate → acetaldehyde	2	- 0.20
NADP ⁺ + H ⁺ + 2 e ⁻ → NADPH	2	- 0.32
NAD ⁺ + H ⁺ + 2 e ⁻ → NADH	2	- 0.32
2 H ⁺ + 2 e ⁻ → H ₂ (g) (pH = 7)	2	- 0.41
acetate → acetaldehyde	2	- 0.60
α-ketoglutarate → succinate + CO ₂ (g)	2	- 0.67



$$(\Delta G^{\circ'} = -nFE^{\circ'}_{\text{el}} = -2 \cdot 96500 \cdot 0.13 = -25090 \text{ J})$$



redox process (ox + n e ⁻ → red)	n	ε ^{o'} (V)
$\frac{1}{2} \text{O}_2 (g) + 2 \text{H}^+ + 2 \text{e}^- \rightarrow \text{H}_2\text{O} (l)$	2	0.81
cytochrome-a ₃ (Fe ³⁺) + e ⁻ → cytochrome-a ₃ (Fe ²⁺)	1	0.55
cytochrome-a (Fe ³⁺) + e ⁻ → cytochrome-a (Fe ²⁺)	1	0.29
cytochrome-c (Fe ³⁺) → cytochrome-c (Fe ²⁺)	1	0.25
cytochrome-c ₁ (Fe ³⁺) → cytochrome-c ₁ (Fe ²⁺)	1	0.22
cytochrome-b (Fe ³⁺) → cytochrome-b (Fe ²⁺)	1	0.07
ubiquinone + 2H ⁺ + 2e ⁻ → ubiquinol	2	0.04
NADH dehydrogenase (FMN) + 2H ⁺ + 2e ⁻ → NADH dehydrogenase (FMNH ₂)	2	-0.03
NADP ⁺ + H ⁺ + 2 e ⁻ → NADPH	2	-0.32
NAD ⁺ + H ⁺ + 2 e ⁻ → NADH	2	-0.32
2 H ⁺ + 2 e ⁻ → H ₂ (g) (pH = 7)	2	-0.41

Question

Put the following components of the respiratory chain in correct order as the electrons flow in the mitochondrion!

- a) Cytochrome c ($\text{Fe}^{3+}/\text{Fe}^{2+}$) $\epsilon^{\circ} = + 0.26 \text{ V}$
- b) Q / QH_2 $\epsilon^{\circ} = - 0.10 \text{ V}$
- c) Cytochrome c_1 ($\text{Fe}^{3+}/\text{Fe}^{2+}$) $\epsilon^{\circ} = + 0.23 \text{ V}$
- d) NAD^+/NADH $\epsilon^{\circ} = - 0.32 \text{ V}$
- e) Cytochrome b ($\text{Fe}^{3+}/\text{Fe}^{2+}$) $\epsilon^{\circ} = + 0.04 \text{ V}$